

IN THE CLAIMS:

This listing of claims will replace all prior versions, and listing, of claims in the application.

Listing of the Claims:

1. (Currently amended) A method of histological assessment of nuclear pleomorphism by identifying image regions comprising respective groups of contiguous pixels and potentially corresponding to cell nuclei in histological image data, the method including using computer apparatus to carry out the steps of:
 - a) thresholding the image data to render it binary with identified image regions and background distinguished from one another by association with different binary digits,
 - b) determining perimeters and areas of identified image regions,
 - c) calculating image region shape factors from the perimeters and areas, and
 - d) assessing nuclear pleomorphism from the shape factors' statistical parameters.
2. (Previously presented) A method according to Claim 1 wherein the shape factors' statistical parameters comprise at least one of their mean, weighted mean, median, mode, maximum and minimum.
3. (Previously presented) A method according to Claim 1 wherein the step of thresholding the imaged data is Otsu thresholding.
4. (Currently amended) A method of histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell

nuclei in histological image data, the method including using computer apparatus to carry out the steps of:

- a) thresholding the image data to render it binary,
- b) determining perimeters and areas of identified image regions,
- c) calculating image region shape factors from the perimeters and areas, and
- d) assessing nuclear pleomorphism as being relatively low, moderate or high according to whether the mean or median of the shape factors is relatively low, moderate or high respectively.

5. (Previously presented) A method according to Claim 4 wherein a shape factor S for an image region potentially corresponding to a cell nucleus is given by
$$S = \frac{kP^2}{A}$$
, where k is a constant, P is image region perimeter and A is image region area, and a mean shape factor S_m for a set of image regions potentially corresponding to cell nuclei is thresholded as $S_m \leq 30k$ (low), $30k < S_m \leq 35k$ (moderate) and $S_m > 35k$ (high) corresponding to pleomorphism being relatively low, moderate or high respectively.

6. (Previously presented) A method according to Claim 1 wherein the step of thresholding the image data to render it binary is preceded by transforming colour image data into greyscale image data with improved image definition compared to an individual red green or blue plane in colour image data, and the step of thresholding the image data is carried out upon the greyscale image data.

7. (Previously presented) A method according to Claim 6 wherein the step of transforming colour image data into greyscale image data is carried out by Principal Component Analysis (PCA) in which the greyscale image data is a first principal component.

8. (Currently amended) A method of histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological colour image data, the method including using computer apparatus to carry out the steps of:

- a) decomposing colour image data into a set of sub-images each of which overlaps half of each of its neighbours,
- b) applying Principal Component Analysis (PCA) to each sub-image to obtain greyscale sub-images as first principal components, in order to provide improved image definition compared to an individual red green or blue plane in the colour image data,
- c) removing from each sub-image regions at sub-image edges potentially corresponding to cell nuclei,
- d) thresholding the greyscale image data to render it binary, and
- e) determining perimeters and areas of identified image regions, calculating image region shape factors from the perimeters and areas and assessing nuclear pleomorphism from the shape factors' statistical parameters.

9. (Currently amended) A method of histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological image data, the method including using computer apparatus to carry out the steps of:

- a) filtering the image data to overwrite regions which are not of interest using a filtering process which does not appreciably affect image region perimeter,
- b) thresholding the image data to render it binary,
- c) determining perimeters and areas of identified image regions,
- d) calculating image region shape factors from the perimeters and areas, and
- e) assessing nuclear pleomorphism from the shape factors' statistical parameters.

10. (Previously presented) A method according to Claim 9 wherein the step of overwriting regions which are not of interest includes setting relatively small image regions to a background pixel value and setting hole pixels in relatively larger image regions to a non-hole image region pixel value.
11. (Currently amended) A method of histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological image data, the method including using computer apparatus to carry out the steps of:
 - a) dividing the image data into overlapping sub-images, applying PCA to each sub-image to provide a respective greyscale sub-image and removing from the greyscale sub-images:
 - i) image regions touching or intersecting sub-image boundaries,
 - ii) unsuitably small image regions, and
 - iii) holes in relatively large image regions,
 - b) reassembling the sub-images into a binary image by thresholding,
 - c) determining perimeters and areas of identified image regions, and
 - d) calculating image region shape factors from the perimeters and areas and assessing nuclear pleomorphism from the shape factors' statistical parameters.
12. (Previously presented) Apparatus for histological assessment of nuclear pleomorphism by identifying image regions comprising respective groups of contiguous pixels and potentially corresponding to cell nuclei in histological image data, the apparatus incorporating a computer programmed to threshold the image data to render it binary with identified image regions and background distinguished from one another by association with different binary digits, determine perimeters and areas of identified image regions, calculate image region shape factors from the perimeters and areas and assess nuclear pleomorphism from the shape factors' statistical parameters.

13. (Previously presented) Apparatus according to Claim 12 wherein the shape factors' statistical parameters comprise at least one of their mean, weighted mean, median, mode, maximum and minimum.
14. (Previously presented) Apparatus according to Claim 12 wherein the computer is programmed to threshold the image data using Otsu thresholding.
15. (Previously presented) Apparatus for histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological image data, the apparatus incorporating a computer programmed to threshold the image data to render it binary, determine perimeters and areas of identified image regions, calculate image region shape factors from the perimeters and areas and to assess nuclear pleomorphism as being relatively low, moderate or high according to whether the mean or median of the shape factors is relatively low, moderate or high respectively.
16. (Previously presented) Apparatus according to Claim 15 wherein the computer is programmed to determine a shape factor S for an image region potentially corresponding to a cell nucleus is given by $S = \frac{kP^2}{A}$, where k is a constant, P is image region perimeter and A is image region area, and the computer is also programmed to determine a mean shape factor S_m for a set of image regions potentially corresponding to cell nuclei, to threshold the mean shape factor as $S_m \leq 30k$ (low), $30k < S_m \leq 35k$ (moderate) and $S_m > 35k$ (high) and to indicate pleomorphism being relatively low, moderate or high respectively.
17. (Previously presented) Apparatus according to Claim 12 wherein the computer is programmed to implement a transformation of colour image data into greyscale image data with improved image definition compared to an individual red green or blue plane in colour image data, and to implement such transformation prior to thresholding the image data to render it binary, and the computer is also

programmed to carry out thresholding of the image data using the greyscale image data.

18. (Previously presented) Apparatus according to Claim 17 wherein the computer is programmed to transform colour image data into greyscale image data using PCA in which the greyscale image data is a first principal component.
19. (Previously presented) Apparatus for histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological colour image data, the apparatus incorporating a computer is programmed to:
 - a) divide the colour image data into overlapping sub-images,
 - b) apply Principal Component Analysis (PCA) to the sub-images to obtain greyscale sub-images as first principal components, in order to provide improved image definition compared to an individual red green or blue plane in the colour image data,
 - c) remove from the greyscale sub-images:
 - i) image regions touching or intersecting sub-image boundaries,
 - ii) unsuitably small image regions, and
 - iii) holes in relatively large image regions,
 - d) reassemble the sub-images into a binary image by thresholding,
 - e) determine perimeters and areas of identified image regions, and
 - f) calculate image region shape factors from the perimeters and areas and assessing nuclear pleomorphism from the shape factors' statistical parameters.
20. (Previously presented) Apparatus for histological assessment of nuclear pleomorphism by identifying image regions potentially corresponding to cell nuclei in histological image data, the apparatus incorporating a computer is programmed to threshold the image data to render it binary, set relatively small image regions to a background pixel value and to set hole pixels in relatively

larger identified image regions to a non-hole image region pixel value, determine perimeters and areas of identified image regions, calculate image region shape factors from the perimeters and areas and assess nuclear pleomorphism from the shape factors' statistical parameters.

21. (Previously presented) A computer software product comprising a computer readable hardware medium containing computer readable instructions for use in histological assessment of nuclear pleomorphism for controlling a computer to identify image regions comprising respective groups of contiguous pixels and potentially corresponding to cell nuclei in histological image data, the software product also having computer readable instructions for controlling a computer to threshold the image data to render it binary with identified image regions and background distinguished from one another by association with different binary digits, determine perimeters and areas of identified image regions, calculating image region shape factors from the perimeters and areas and assess nuclear pleomorphism from the shape factors' statistical parameters.
22. (Previously presented) A computer software product according to Claim 21 wherein the shape factors' statistical parameters comprise at least one of their mean, weighted mean, median, mode, maximum and minimum.
23. (Previously presented) A computer software product according to Claim 21 wherein the computer readable instructions include instructions for controlling a computer to threshold the imaged data using Otsu thresholding.
24. (Previously presented) A computer software product comprising a computer readable medium containing computer readable instructions for use in histological assessment of nuclear pleomorphism for controlling a computer to identify image regions potentially corresponding to cell nuclei in histological image data, wherein the computer readable instructions being for controlling a computer to determine nuclear pleomorphism as being relatively low, moderate

or high according to whether the mean or median of the shape factors is relatively low, moderate or high respectively.

25. (Previously presented) A computer software product according to Claim 24 wherein the computer readable instructions are also for controlling a computer to:
 - a) determine a shape factor S for an image region potentially corresponding to a cell nucleus given by $S = \frac{kP^2}{A}$, where k is a constant, P is image region perimeter and A is image region area,
 - b) threshold a mean shape factor S_m for a set of image regions potentially corresponding to cell nuclei as $S_m \leq 30k$ (low), $30k < S_m \leq 35k$ (moderate) and $S_m > 35k$ (high), and
 - c) to indicate nuclear pleomorphism being relatively low, moderate or high respectively.
26. (Previously presented) A computer software product according to Claim 21 wherein the computer readable instructions are also for controlling a computer so that before thresholding the image data to render it binary such computer will transform colour image data into greyscale image data with improved image definition compared to an individual red green or blue plane in colour image data, and subsequently such computer will implement thresholding of the image data using the greyscale image data.
27. (Previously presented) A computer software product according to Claim 26 wherein the computer readable instructions are also for controlling a computer to transform colour image data into greyscale image data by PCA in which the greyscale image data is a first principal component.
28. (Previously presented) A computer software product comprising a computer readable medium containing computer readable instructions for use in histological assessment of nuclear pleomorphism by identifying image regions

potentially corresponding to cell nuclei in histological image data, the computer readable instructions being for controlling a computer to:

- a) decompose colour image data into a set of sub-images each of which overlaps half of each of its neighbours,
- b) apply Principal Component Analysis (PCA) to each sub-image to obtain greyscale sub-images as first principal components, in order to provide improved image definition compared to an individual red green or blue plane in the colour image data,
- c) remove from each greyscale sub-image:
 - i) image regions touching or intersecting sub-image boundaries,
 - ii) unsuitably small image regions, and
 - iii) holes in relatively large image regions,
- d) reassemble the sub-images into a binary image by thresholding,
- e) determine perimeters and areas of identified image regions, and
- f) calculate image region shape factors from the perimeters and areas and assessing nuclear pleomorphism from the shape factors' statistical parameters.

29. (Previously presented) A computer software product according to Claim 21 wherein the computer readable instructions are also for setting relatively small image regions to a background pixel value and for setting hole pixels in relatively larger image regions to a non-hole image region pixel value.